

# Integration of demography and genetics in population restorations<sup>1</sup>

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**Abstract:** By definition, restoration projects involve small populations, which are subject to demographic, genetic, and environmental stochasticities. Besides its fundamental interest for ecology, the integration of these stochastic factors in population viability models is required to make such models more realistic. In this paper, we report previous attempts to integrate population dynamics and population genetics, two disciplines that are generally treated as separate fields. We then evaluate their potential interactions in the context of population restoration. In the first part, we investigate the interactions among stochastic factors of extinction and various species characteristics, such as growth rate, generation length, and mating system. In the second part, we discuss how demographic and genetic models can be used to compare the relative efficiencies of different reintroduction and reinforcement strategies in different environments. For that purpose, we examine spatial and temporal aspects of release, as well as the number and type of individuals to release. In many cases, these comparisons uncover opposite effects among the genetic and demographic factors that antagonistically influence short- and long-term viabilities. Choosing an appropriate restoration strategy should therefore involve the integration of different disciplines in population viability analyses.

**Keywords:** conservation genetics, demographic stochasticity, inbreeding depression, population dynamics, population viability analysis, restoration strategy.

**Résumé :** Par définition, les projets de restauration impliquent de petites populations soumises à des stochasticités démographique, génétique et environnementale. En plus de leur intérêt fondamental en écologie, l'intégration de ces facteurs stochastiques dans les modèles de viabilité des populations est nécessaire afin de rendre les modèles plus réalistes. Dans cet article nous décrivons de précédentes tentatives d'intégrer la dynamique des populations et la génétique des populations, deux disciplines normalement traitées séparément. Nous évaluons ensuite leurs interactions potentielles dans le contexte de la restauration de populations. Dans une première partie, nous étudions les interactions entre les facteurs stochastiques d'extinction et diverses caractéristiques des espèces telles que le taux de croissance, la durée de génération et le système de reproduction. Dans une deuxième partie, nous discutons comment les modèles démographique et génétique peuvent être utilisés pour comparer l'efficacité relative de différentes stratégies de réintroduction et de renforcement et ce dans différents environnements. Dans ce but, nous examinons les aspects spatiaux et temporels du lâcher ainsi que le nombre et le type d'individus à libérer. Dans plusieurs cas, ces comparaisons font ressortir des effets opposés des facteurs génétiques et démographiques qui influencent de façon antagoniste la viabilité à court et à long terme. Le choix d'une stratégie de restauration adéquate devrait donc impliquer l'intégration de différentes disciplines dans des analyses de viabilité des populations.

**Mots-clés :** analyse de viabilité des populations, dépression de consanguinité, dynamique des populations, génétique de la conservation, stochasticté démographique, stratégie de restauration.

**Nomenclature:** Throughout, latin binomials are those used by the original authors.

## Introduction

Reintroductions and reinforcements, which may involve wild-caught or captive-bred animals, have become an increasingly important tool for maintenance of demographically and genetically viable populations (Beissinger & McCullough, 2002; Morris & Doak, 2002). Following the definitions given by the World Conservation Union, the term *reintroduction* refers to an attempt to establish a species in an area that was once part of its historical range but from which it has become extinct, while a *reinforcement* is the addition of individuals to an existing population of conspecifics (IUCN, 1998). Both methods may involve *translocations* (the mediated movement of wild individu-

als or populations from one part of their range to another). Numerous studies have investigated the feasibility and efficiency of different translocation methods using empirical pre-existing data (Griffith *et al.*, 1989; Van Dierendonck & Wallis de Vries, 1996; Wolf *et al.*, 1996; Green, 1997), experimentation (Bright & Morris, 1994; Castro *et al.*, 1994), as well as demographic (McCarthy, 1994; Novellie, Millar & Lloyd, 1996; Legendre *et al.*, 1999; Sarrazin & Legendre, 2000) and genetic models (Hedrick, 1995; Earnhardt, 1999), and the value of restoration as a test for basic ecological theory has been previously highlighted (Jordan, Gilpin & Aber, 1987; Sarrazin & Barbault, 1996; Montalvo *et al.*, 1997).

Although they exhibit some peculiarities, restored populations face generally the same threats as natural ones. In particular, the demographic and genetic problems involved in the small population paradigm (*sensu*

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Caughley, 1994) have been widely discussed in fundamental population biology as well as in applied conservation biology. However, despite the consensus that a multidisciplinary approach is the most efficient in conservation sciences (Soulé 1985), these disciplines are generally treated as separate fields (Clarke & Young, 2000; Oostermeijer, Luijten & den Nijs, 2003).

One of the common characteristics of most restoration projects is that they generally involve, at least temporarily, small populations. Demographic stochasticity (*i.e.*, the variance in population growth caused by chance realizations of individual probabilities of death and birth events) is frequently invoked as an important cause of extinction in small populations (Leigh, 1981) and may synergistically interact with environmental stochasticity (*i.e.*, variability in population growth rates due to changes in birth and death rates from one season to the next in response to environmental factors). Small population size is also one of the most important sources of genetic problems. The detrimental effects of small population size differ according to whether it is brief (bottleneck), owing to a small number of released founders, or durable, owing to a low growth rate or limited carrying capacity. In the former case, the major genetic threat is associated with the rapid increase of the frequency of individuals that are homozygous for deleterious alleles identical by descent, resulting in the reduction of fitness termed inbreeding depression (see Hedrick & Kalinowski, 2000 for review and discussion of recent studies on inbreeding depression). In the latter case, gradual processes will lead to an increase of the frequency of deleterious mutations present in the population (Lynch, Conery & Bürger, 1995) and to the progressive loss of genetic variation necessary for adaptation to environmental changes (Reed *et al.*, 2003a). Such variations in allelic frequencies due to drift and inbreeding are sometimes referred to as *genetic stochasticity*.

Supplementing our theoretical understanding of deleterious genetic processes occurring in small isolated populations (Frankham, Ballou & Briscoe, 2002; Hedrick, 2004), analyses of extensive data sets have uncovered strong relationships between population size, genetic diversity, and fitness (Reed & Frankham, 2003; Reed, 2005), and recent studies have provided strong evidence of the effect of inbreeding on extinction risk (*e.g.*, Spielman, Brook & Frankham, 2004).

The genetic and demographic peculiarities of artificially reintroduced populations are generally related to the restoration protocol. For instance, genetic diversity may be very low in cases where founders originate from one captive stock or very high in cases where founders derive from various populations (Tordoff & Redig, 2001). In both cases, specific problems may be encountered. Reintroduced or reinforced populations may also exhibit a demographic disequilibrium (*e.g.*, in terms of sex or age structure), strongly affecting their dynamics. Hence, specific and realistic modelling of population dynamics is not only important to assess the “purely” demographic threats, but also because demographic parameters, life history traits, and initial conditions strongly influence short- and long-term genetic processes. In the first part of this paper, we assess the effect of various species characteristics, such as growth rate, genera-

tion length, and mating system, on demographic and genetic factors of extinction and on their interactions. In the second part, we examine some situations in which demographic and genetic approaches that account for these species peculiarities can be used to assess the effectiveness of different reintroduction and reinforcement strategies.

### Influence of species characteristics on demographic and genetic mechanisms responsible for population extinction

As mentioned above, the extent of inbreeding is mostly dependent on population size. In order to compare species that have different life history traits, population geneticists use the concept of effective size ( $N_e$ ), which corresponds to the number of individuals that would result in the same genetic drift in an idealized population. Effective size is mostly determined by the real (census) past and present sizes of the population, which depend themselves on number of founders, maximum growth rate, carrying capacity, and any extrinsic factors that lead to temporal variation in size. Additionally, for a given census size  $N$ ,  $N_e$  may also vary with variance in lifetime reproductive success, which depends on many demographic characteristics of the population, such as sex-ratio, social structure, and mating system.

#### RATE OF INCREASE AND POPULATION SIZE

Although inbreeding is mostly examined in laboratory conditions in *Drosophila* species (Latter *et al.*, 1995; Bijlsma, Bundgaard & Boerema, 2000), empirical support for its negative effects has been recorded for various taxa in nature (review in Keller & Waller, 2002) and captivity (Ralls, Ballou & Templeton, 1988; Ballou, 1997). Although some within-species variation has been reported, little variation among major taxonomic groups has been found in susceptibility to inbreeding (Ralls, Ballou & Templeton, 1988; Crnokrak & Roff, 1999). Different studies based on realistic population viability analyses found contrasting effects of inbreeding depression (see for instance Burgman & Lamont, 1992 and Oostermeijer, 2000). In a recent study, Brook *et al.* (2002) conducted population viability analyses of several threatened species to investigate the impact of inbreeding depression on viability. Although the impacts were similar across major taxa, they uncovered some differences among species due to different initial growth rates.

The short-term interactions between inbreeding depression and population growth rate and their effect on population persistence can be illustrated using a model integrating demographic and genetic processes:

The rate of replacement of the population at generation  $t$  is given by

$$R_t = R_0 e^{-Bf_t}$$

where  $R_0$  is the basic replacement rate (*i.e.*, in the absence of inbreeding depression),  $B$  represents the lethal equivalents (*i.e.*, the extent of the negative effects of inbreeding), and  $f_t$  quantifies the population inbreeding at generation  $t$ , given by

$$f_{t+1} = 1/2N_e + (1 - 1/2N_e)f_t \text{ (Falconer, 1989).}$$

The effective population size at generation  $t$  is therefore given by

$$N_{e,t+1} = R_t N_e$$

Figure 1 presents the effects of  $R_0$  and of the initial effective population size  $N_{e0}$  on population growth in a context of reintroduction, comparing a situation with a low number of founders and a high replacement rate (Figure 1,  $N_0 = 2$ ) and a situation with a larger number of founders and a lower replacement rate (Figure 1,  $N_0 = 10$ ). Although inbreeding increases more during the first generations in population 2 due to the small initial size, the effects of inbreeding depression (here with  $B = 1$  lethal equivalent) on  $N_{e_t}$  are qualitatively more detrimental in population 10 for 2 reasons: 1) in population 2, population size increases rapidly, so inbreeding significantly increases only during the very first generations, and 2) in population 2, owing to the high value of  $R_0$ ,  $R_t$  remains higher than 1 even for a strong inbreeding depression, which allows population size to increase longer. As with previous theoretical results (Mills & Smouse, 1994), this simple illustration shows that the short-term effects of inbreeding are more harmful in slow-growing species. Starting from 2 individuals does not necessarily lead to population extinction, even though inbreeding depression might be large. Success of small populations does not mean that inbreeding depression does not impair population success in the wild, but rather that population growth is large enough such that inbreeding does not accumulate in the population concerned.

In the short-term effects described above, inbreeding depression was assumed to remain constant. In reality, due to purging effects or accumulation of deleterious alleles, it may vary along generations. Further, inbreeding depression is typically measured as an intra-population relative index (*i.e.*, by comparing the fitness of inbred *versus* non-inbred individuals in the same population). However, in the long run, deleterious genetic changes may affect all individuals in the population. Assessing these longer-term effects requires us to explicitly take into account gradual genetic changes in a given demographic context (population growth and regulation).

From a demographic viewpoint, in the absence of any mechanism of regulation, a population will either increase or decrease. In the former case, even with a small number of founders, population will be small only for a brief period of time. In the latter case, population is fated to extinction. However, in the long run, population size is mostly determined by density-dependent processes affecting its growth. Density dependence may play a critical role in restored populations, particularly in cases where the species habitat is restricted. There is an important disparity of population sizes among the different endangered managed taxa. For instance, herbivore species exhibit generally large population sizes, while carnivore populations frequently comprise fewer than 1000 individuals (Goodman, 1987). The effective size of these populations is generally even smaller, as  $N_e$  may be only a fraction of the total population of adults (a review of published estimates suggests that the average ratio of  $N_e$  to adult numbers is 0.1; Frankham, 1995). From a genetic viewpoint, for such effective population sizes, mildly deleterious mutations will tend to become fixed and accumulate (because selection is less efficient in small populations), progressively increasing the genetic load of the population (*i.e.*, the difference in fitness between the popu-

lation and a hypothetical population composed solely of the fittest genotype). Unlike inbreeding depression, this process involves a gradual change in the frequency of alleles and requires a longer period of time to be effective. However, as in the case described above, the gradual reduction of fitness ultimately leads to a decrease of population size. This facilitates further mutation accumulation by enhanced genetic drift, leading to a “mutational meltdown” (Lynch, Conery & Bürger, 1995; Whitlock, 2000). The short- and long-term effects of inbreeding as a function of  $N_e$  are summarized in Table I.

GENERATION TIME

During the late 1980s and 1990s the contributions of inbreeding depression and mutation accumulation to population extinction were theoretically and experimentally examined (Lande, 1994; Gilligan *et al.*, 1997) and discussed (Lande, 1988; Caughley, 1994). These studies led some authors to the conclusion that genetic factors had only a minor impact on species/population extinction compared with demographic and environmental threats. Although most of the papers cited above are controversial (Hedrick *et al.*, 1996, Lynch *et al.*, 1999), they emphasize the importance of the time scales considered in conservation. As underlined by Gilligan *et al.* (1997), typical time frames of concern for captive propagation programs are 100–200 y. This duration

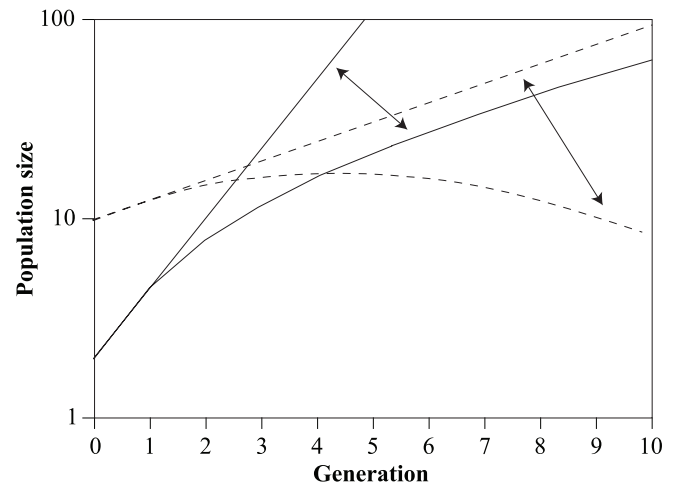


FIGURE 1. Founding population dynamics: influences of  $R_0$  and  $N_0$ . Solid line:  $R_0 = 2.25$  and  $N_0 = 2$ ; dashed line:  $R_0 = 1.25$  and  $N_0 = 10$ . The upper curve indicates population size in the absence of inbreeding depression. The lower curve indicates population size in the presence of inbreeding depression ( $B = 1$ ).

TABLE I. Decrease of population mean fitness as a function of the effective size  $N_e$  ( $R$  is the basic replacement rate of the population).

Population size	Relative decrease of vital rates per generation	
	Short-term $\sim 1/2N_e$	Long-term $\sim 1/20N_e$
$N_e < 50$	$> 1\%$	$> 1/1000$ (Extinction time $< 1000R$ generations)
$50 < N_e < 1000$	$< 1\%$	$> 1/20\,000$ (Extinction time $< 20\,000R$ generations)

represents 2000 to 5000 generations for *Drosophila* species in laboratory conditions. However, it typically represents fewer than 50 generations for species of conservation concern. Since genetic processes operate on a per generation basis (Falconer, 1989), population geneticists use the generation as a time unit, while ecologists and managers use absolute time measures (*e.g.*, year). This apparent dissociation between genetics and demography can partially be solved using the generation time ( $T$ ), computed as the average age of parents giving birth in a population in stable age distribution (Caswell, 2001). In practice, the generation time enables calculation of the number of generations elapsed for a given period of time. Using this approach makes it possible to compare small populations of different species (for instance a short-lived *versus* a long-lived one). For the same absolute duration, gradual genetic processes are expected to be more detrimental in a short-lived species compared with a long-lived one, due to the larger number of generations elapsed. However, this does not mean that the relative contribution of genetic factors to population extinction is higher in such species compared with non-genetic factors. Indeed, some environmental and demographic aspects related to generation length may scale with the genetic aspects. From a genetic view-point, using data from several taxa, Lynch *et al.* (1999) showed that the average mutational heritabilities and the mutational coefficients of variation were higher for species with longer generation times, which are expected to have greater numbers of germline cell divisions. This result is consistent with the observations of Drake *et al.* (1998), suggesting that the number of mutations per effective genome per cell division is approximately constant across a diversity of eukaryotic species. Species of conservation concern with complex genomes, a high number of germline cell divisions, and long generation times are likely to have higher mutation rates per generation than laboratory species (such as *Drosophila* and nematode species), from which mutation estimates derive.

From a demographic viewpoint, short-lived species typically exhibit a higher annual growth rate ( $R$ ) than long-lived ones, but this does not mean that the rate of increase per generation is different (Niel & Lebreton, 2005). For instance, the rate for a short-lived species with  $R = 1.15$  and  $T = 3$  and the rate for a long-lived species with  $R = 1.05$  and  $T = 9$  are equivalent. Another factor that obscures the effect of generation time is that demographic stochasticity has a stronger impact on short-lived species, due to a rapid turnover, which amplifies the stochastic fluctuations in the number of offspring (Legendre *et al.*, 1999). Such species therefore have a lower translocation success compared with long-lived ones (Griffith *et al.*, 1989). The frequency of extreme environmental perturbations has also been shown to be strongly related to the generation length, with perturbations being more frequent in short-lived than in long-lived species (Reed *et al.*, 2003b).

Many insights into evolutionary genetics have come from detailed laboratory studies in *Drosophila*. Most *Drosophila* studies investigating the effect of spontaneous mutations on life history traits focus on lifetime fitness, measured as individual fitness components such as mortality rates (Lynch, 1985; Pletcher, Houle & Curtsinger,

1998), longevity (Houle *et al.*, 1994), fecundity, or productivity (Lynch, 1985; Houle *et al.*, 1994). In other species, the impact of deleterious mutations is generally measured on a single trait. The cost of inbreeding in captive mammal populations has been documented using the regression of juvenile survival on the inbreeding coefficient (Ralls, Ballou & Templeton, 1988). In natural conditions, individual survival ability (Keller *et al.*, 1994; Coltman *et al.*, 1999) or fecundity parameters (Bouzat *et al.*, 1998) have been used as fitness indicators. If deleterious genetic factors are expressed in terms of effect on a single demographic rate, their expected impact on population dynamics should be strongly dependent on the demography of the species. The sensitivity of the growth rate of a population to a reduction in a given demographic parameter may indeed vary according to the type of life cycle considered (Caswell, 2001). For instance, long-lived bird species (such as Charadriiformes) generally exhibit high adult survival rates and low annual fecundities. In such species, the population growth rate is primarily sensitive to adult survival. In contrast, populations of short-lived species with low survival rates and high fecundities (such as Passeriforme species) are particularly affected by a decrease of fecundity (Ferrière *et al.*, 1996; Saether & Bakke, 2000). Considering some detrimental effects of the genetic load on a single demographic component will therefore not have the same impact on population viability for short- *versus* long-lived species.

#### REPRODUCTIVE STRATEGY

The type of reproductive strategy plays a major role both in genetic and demographic mechanisms. Its role is particularly complex in plants, in which sexual (including outcrossing, selfing, or a combination of both) and asexual reproduction are found. Additionally, the cost of inbreeding depends on a variety of mechanisms, such as polyploidy, genetically controlled self-incompatibility systems, biased movements of pollen and seed, and complex interspecific interactions with pollinators. Selfing species typically suffer less inbreeding depression than outbreeding species, due to recurrent purging (Lande & Schemske, 1985; Husband & Schemske, 1996). Short-term effects of inbreeding (see above) are consequently more dramatic for outcrossing species. In contrast, long-term effects of inbreeding are identical for both types of species, because the rate of fixation of deleterious alleles is identical (Charlesworth, 1992). However, variation in the reproductive strategy as a function of population size might reduce the validity of estimates of the impact of inbreeding. In particular, some species show dramatic reduction in cross-fertilization rates in small populations (Raijmann *et al.*, 1994). Since selfing species have less heterozygosity and more differentiation among populations than outbreeders (Liu, Zhang & Charlesworth, 1998), priority must be given to the restoration of distinct connected populations for such species. Heterosis, and thus the advantage of restoring the connection, generally increases with the selfing rate (Theodorou & Couvet, 2002).

In contrast to plant species, where a variety of physiological subtleties exist, behavioural complexities (*e.g.*, sexual selection) in animal mating systems are expected to strongly influence the genetic and demographic processes involved in extinction. This impact of the mating system

can be illustrated by a comparison between a polygamous and a monogamous population (Figure 2). The impact of demographic stochasticity is stronger for monogamous populations compared with polygamous ones (Legendre *et al.*, 1999), which engenders a short-term advantage for polygamous populations. On the other hand, because males contribute more equally to reproduction compared with polygyny, monogamy is advantageous from a genetic viewpoint. This induces a long-term advantage for monogamous species.

### Restoration strategies

#### HOW MANY INDIVIDUALS SHOULD BE INTRODUCED? OPTIMAL NUMBER

One particularity of reintroduction projects is that they may involve the release of a small number of individuals taken from large (natural) populations. This implies a precipitous reduction of population size, which tends to create strong inbreeding depression because selection has not had the opportunity to rid the population of deleterious alleles (Soulé, 1980; Lande, 1988). In addition, small populations are extremely vulnerable to demographic stochasticity (Shaffer, 1987). Demographic and genetic theories both predict that the persistence time of a population increases with its initial size. This obvious prediction is consistent with empirical observations suggesting that reintroduction success strongly increases with the number of released founders (Griffith *et al.*, 1989; Green, 1997). However, it is clear that the critical (that is, below which the probability of success is very low) number of founders required will vary among taxa, depending on the life history strategy of the species considered. In particular, this critical number increases for species with a low growth rate (see Figure 1). Hence, no universal rule can be provided concerning the number of individuals to release. The only general observation is that the impact of demographic stochasticity varies with generation length. Releases of fewer than 100 individuals are generally sufficient to ensure short-term population persistence in long-lived species, while 2 to 3 times more are necessary for short-lived ones (Legendre *et al.*, 1999). In many cases, there are additional genetic factors that should

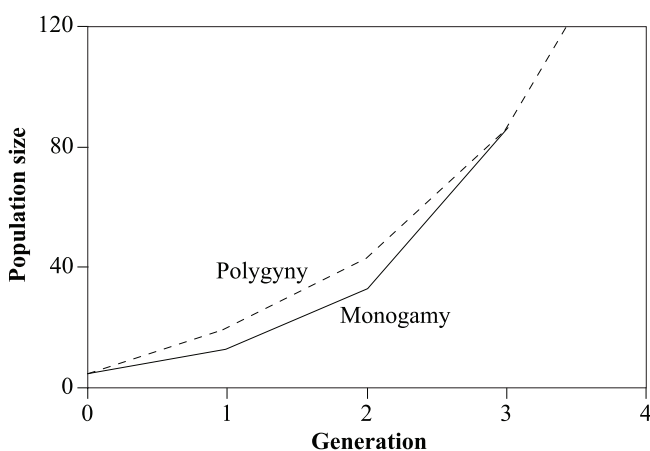


FIGURE 2. Founding population dynamics: influence of the mating system.  $R_0 = 4$ ;  $N_0 = 5$ ;  $B = 1$ ; for polygyny: harem size = 5.

not be neglected. For instance, a second bottleneck can have a very negative effect when the first bottleneck was severe (Thévenon & Couvet, 2002), a fact to take into account when reintroduced individuals derive from a captive population that originated from a limited number of individuals.

In the case of small remnant populations threatened by demographic and environmental uncertainties, it has been theoretically demonstrated that managed translocations of 1 to 6 individuals per year may strongly enhance the probability of species persistence and reduce the required size of nature reserves (Lubow, 1996). From a genetic viewpoint, the number of translocated individuals involved in reinforcements determines the gene flow necessary to maintain genetic variability. Recent studies suggest that an influx on the order of 1 migrant per generation may not be sufficient to impede mutation accumulation in small populations (Couvet, 2002). Conversely, due to the deleterious impact of gene flow on local adaptation, a maximum number of 10 migrants per generation has been proposed (Mills & Allendorf, 1996). Thus, from a genetic viewpoint, the optimal number of translocated individuals should be chosen in order to engender an intermediate gene flow (between 1 and 10 migrants per generation).

#### WHERE INDIVIDUALS SHOULD BE RELEASED: SPATIAL RELEASE STRATEGY

The heterogeneity of environmental conditions is expected to engender heterogeneity among populations of the same species with respect to their provenance. This heterogeneity is important in restoration projects, which frequently involve long-distance translocations. The home-site advantage hypothesis, which predicts that the relative success of introduced populations will decrease as their genetic and environmental distance to the local native population increases, is supported by experimental studies (Montalvo & Ellstrand, 2000; Keller, Kollmann & Edwards, 2000) and empirical data on restorations (Griffith *et al.*, 1989). However, geographic distance is insufficient to predict the extent of genetic differentiation among populations and the importance of local adaptation (Montalvo & Ellstrand, 2000), and the relative contribution of this phenomenon to viability varies among populations, depending on local conditions (*e.g.*, their temporal and spatial scales of variation) (discussion in Wilkinson, 2001). In the case where individuals are translocated to a distant remnant population, hybridization may bring further threats. In particular, outbreeding depression (reduced fitness in hybrid offspring) can occur for environmental (loss of local adaptation) and physiological (loss of intrinsic co-adaptation) reasons (Templeton, 1986). Hence, priority must be given to reinforcement strategies involving locally adapted individuals, if such individuals exist.

In the context of populations released in fragmented habitats, the question of spatial release strategy (*i.e.*, multi-patch *versus* one-patch release) is crucial. From a demographic viewpoint, the one-patch release strategy maximizes initial density, which should minimize Allee effects and demographic stochasticity. However, when reintroducing a large number of individuals into variable environments, the multi-patch release is less risky, at least if local environmental fluctuations (*i.e.*, environmental stochasticity) are

not synchronized among sites (Harrison & Quinn, 1989). The heterogeneity of environmental conditions should also be taken into account for genetic reasons, as it strongly affects the patterns of local adaptation. In the case of a one-patch release, niche conservatism (Holt, 1996) may indeed impede local adaptation to other sites, a hindrance enhanced by demographic stochasticity (Robert, Couvet & Sarrazin, 2003). The benefits of multi-patch introduction have been highlighted for other reasons. It limits, for example, the overall loss of self-incompatibility alleles in self-incompatible plant metapopulations and improves long-term fecundity (Kirchner, Robert & Colas, 2006).

Finally, demographic and genetic theories on the effect of environmental variation on populations suggest that preference should be given to reinforcing or restoring populations with locally adapted individuals, and to releasing them into several connected areas. However, this conclusion is less valid for very small numbers of founders, or when local populations are highly inbred, for which cases priority should be given to maximizing local densities and bringing new genetic variation.

#### WHEN INDIVIDUALS SHOULD BE RELEASED: RATE OF RELEASE

Several studies using demographic population viability assessments in the context of species restoration have concluded that rather than the rate of introduction, the total number of individuals released had the most significant influence on the chance of success (McCarthy, 1994; Legendre *et al.*, 1999; Sarrazin & Legendre, 2000). However, in natural conditions, temporal fluctuations in demographic rates caused by environmental stochasticity tend to increase the efficiency of a restoration with several release events relative to a single release event (Griffith *et al.*, 1989; Haccou & Iwasa, 1996).

Little work has been done on the genetic impact of the rate of introduction on the viability of reintroduced populations. However, simulation models suggest that the best method of release depends on the type of selected genetic variation considered (Robert, Sarrazin & Couvet, 2004a). If deleterious genes are considered, a strategy of staggering release events over a long period of time (multi-event introduction) is advantageous because it engenders a continuous gene flow, which counter-balances the accumulation of deleterious alleles occurring in the resident population (Newman & Tallmon, 2001). In contrast, if adaptive genes are considered, non-adapted newcomers exhibit on average a lower fitness than individuals issued from several generations of local selection (Keller, Kollmann & Edwards, 2000). Their contribution is then dramatically reduced, which impairs the bringing of new adaptive mutations into the population. In such a case, the best strategy is to release all individuals at the same time (one-event introduction). One-event introduction is therefore recommended for restored populations with long-term effective sizes larger than a few hundred individuals (for which genetic deterioration may not pose a serious threat; Whitlock, 2000). In other cases, progressive release should be recommended.

#### WHO SHOULD BE RELEASED: CHOICE OF INDIVIDUALS

The success of species translocations from natural populations into empty sites is believed to be linked to

the genetic characteristics of translocated individuals. As previously mentioned, a major reason for the use of local provenance is the claimed importance of conserving locally adapted genotypes, which are assumed to show high fitness. Such an approach is generally unfeasible, owing to a lack of appropriate individuals, and it involves the risk of producing a population with low genetic variation, precluding future adaptations. The alternative strategy is to release individuals from a large diversity of populations in order to maximize the genetic variability on which selection will act (Tordoff & Redig, 2001; Robert, Couvet & Sarrazin, 2002). This approach may involve the translocation of individuals from large natural (potentially distant) populations. Although it involves the risk of decreasing local adaptation in the local population (Hedrick, 1995; Storfer, 1999), the appropriateness of long-distance reinforcement to declining populations has been demonstrated in several species, such as the Greater Prairie chicken (*Tympanuchus cupido*; Bouzat *et al.*, 1998), the adder (*Vipera berus*, Madsen *et al.*, 1999), and the Florida panther (*Puma concolor coryi*; Hedrick, 2004), in agreement with experimentation (Newman & Tallmon 2001).

When individuals are released from captivity, additional difficulties can further reduce reintroduction success. Captive populations are generally small, and may therefore endure the same genetic deterioration as any small population (*e.g.*, inbreeding depression, accumulation of deleterious mutations, loss of genetic diversity). In addition, some problems specifically related to captivity may arise, such as relaxation of natural selection or adaptation to captivity (Frankham, Ballou & Briscoe, 2002). Thus, the choice of captive individuals used for reintroduction should take into account these possibilities in order to maximize short-term demographic potential (*i.e.*, founders should be healthy, with high reproductive potential) and long-term genetic sustainability (*i.e.*, founders should have high genetic diversity and low adaptation to captivity). This can be achieved by using pedigree information (Ballou, Gilpin & Foose, 1995), as in the example of the California condor (*Gymnogyps californianus*), in which genetic management of captive and restored populations was designed to preserve genetic variation and minimize the effect of chondrodystrophy, a genetically inherited form of dwarfism (Ralls & Ballou, 2004).

Other aspects of the choice of individuals to release concern the sex and age of individuals to release. The optimal sex-ratio of the founding population depends on the reproduction strategy of the species. For instance, from a demographic viewpoint, a female-biased sex ratio may be optimal in polygynous species, although it involves the genetic risks of further decreasing effective population size and/or reducing competition among males for reproduction.

The question of the age of the individuals to release in reintroduction has recently been investigated by Sarrazin and Legendre (2000), who compared 2 alternative methods of release (adult *versus* juvenile releases) using a demographic model applied to the case of a Griffon vulture (*Gyps fulvus*) population reintroduced in southern France. Their results suggest that the release of adults is more efficient than the release of juveniles despite a reduction in survival and fecundity rates in individuals released as adults. This

demographic result holds for both short- and long-lived species. However, other work including genetic considerations suggests that populations founded by adults in the same context may be more affected by accumulation of mutations than those founded by juveniles, leading in some cases to better long-term efficiency when juveniles are released (Robert *et al.*, 2004).

### Conclusion

Although current population dynamics and population genetics theories have enabled conservation biologists to construct a number of qualitative management guidelines for restoration programs, the interactions between demographic and genetic sources of population extinction and various species characteristics prevent them from providing general quantitative recommendations. The development of practical recommendations is complicated by the antagonism between some short-/long-term genetic and demographic effects (*e.g.*, differences in optimal genetic and demographic sex-ratio of released populations) and between different genetic processes (*e.g.*, inbreeding *versus* outbreeding depression).

For reintroduction and reinforcement purposes the quality and number of individuals to release and the spatial and temporal release strategies chosen are highly entangled and should be assessed in a common framework. Although there are few examples of such integrative work, thorough interdisciplinary assessments would certainly improve the success of restoration operations. The genetic restoration of the Florida panther provides an example of such success. Genetic drift fixed deleterious traits in the last population of the subspecies, resulting in extremely low fecundity (Roelke, Martenson & O'Brien, 1993). Theoretical evaluation of reinforcement (Hedrick, 1995) enabled precise recommendations to be made in terms of provenance (other subspecies from Texas), demographic status (young adult females), and number (8) of individuals to release and rate of introduction (20% gene flow from outside in the first generation and 2.5% thereafter). This introduction resulted in substantial improvement of fecundity traits and an increase of population size and range (Hedrick, 2004; Pimm, Dollar & Bass, 2006). Thus, in spite of the difficulties mentioned above, the development of genetic and demographic theories has certainly already contributed to the success of some restoration programs. The integration of these fields in conservation research will certainly benefit future restoration projects when assessed in a given species/population context.

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